of birth, and that the collection of cord blood cells at birth enables one to obtain increased quantities of cells such as CD34+ cells, which are useful in gene therapy when genetically engineered to include a nucleic acid sequence encoding a therapeutic agent. Applicants also are the first ones to demonstrate that one can obtain CD34+ cells from cord blood of a patient, genetically engineer such CD34+ cells to include at least one nucleic acid sequence encoding a therapeutic agent, and return such genetically engineered CD34+ cells to the circulatory system of the patient, whereby the genetically engineered cells express therapeutic amounts of the therapeutic agent *in vivo*. This has been demonstrated in Example 2 of the specification. Although the cited prior art discloses the genetic engineering of CD34+ cells, such prior art does not disclose or even remotely suggest to one of ordinary skill in the art that one may obtain CD34+ cells from cord blood, genetically engineer such CD34+ cells obtained from cord blood and administer such genetically engineered CD34+ cells to a patient to provide a therapeutic effect in such patient. From such cited prior art, one cannot reasonably predict that one may successfully obtain expression of therapeutic amounts of a therapeutic agent *in vivo* from autologous genetically engineered CD34+ cells obtained from cord blood.

In accordance with an aspect of the present invention, there is provided, as defined in Claim 1, a method of providing a therapeutic effect in a human patient. The method comprises administering autologous CD34+ cells obtained from cord blood to the patient. The CD34+ cells have been genetically engineered to include at least one nucleic acid sequence encoding a therapeutic agent. The autologous CD34+ cells are administered in an amount effective to provide the patient with an effective amount of the therapeutic agent by expression of the nucleic acid sequence in the patient.

Another aspect of the invention, as defined in Claim 6, is directed to a method of treating a human patient suffering from severe combined immune deficiency resulting from adenosine deaminase deficiency. In this aspect, the autologous CD34+ cells obtained from cord blood have been genetically engineered to include a nucleic acid sequence encoding adenosine deaminase. The autologous CD34+ cells are administered to the patient in an amount effective to treat severe combined immune deficiency resulting from adenosine deaminase deficiency by providing the patient with an effective amount of the adenosine deaminase by expression of the nucleic acid sequence encoding adenosine deaminase in the patient.

Claim 11 is directed to treating an infant suffering from severe combined immune deficiency which comprises obtaining cord blood from the infant, and separating CD34+ cells from cord blood. The CD34+ cells obtained from cord blood are cultivated in the presence of (i) Interleukin-3; (ii) Interleukin-6; and (iii) a c-kit ligand. The CD34+ cells then are transfected with a nucleic acid sequence encoding adenosine deaminase. The transfected CD34+ cells then are administered to the infant in an amount effective to treat severe combined immune deficiency.

In another aspect of the invention, as defined in Claim 16, there is provided a method of genetically engineering CD34+ cells obtained from cord blood with at least one nucleic acid sequence encoding a therapeutic agent. The method comprises cultivating CD34+ cells obtained from cord blood in the presence of (i) Interleukin-3; (ii) Interleukin-6; and (iii) a c-kit ligand. The CD34+ cells then are transfected with at least one nucleic acid sequence encoding a therapeutic agent.

Anderson discloses the transduction of T-cells with the adenosine deaminase gene. The transduced T-cells then were given to human patients in order to treat severe combined immune deficiency. Although Anderson discloses the possibility of transducing an enriched population of CD34+ cells, Anderson does not disclose or even remotely suggest to one of ordinary skill in the art the transduction of CD34+ cells obtained from cord blood with a nucleic acid sequence encoding a therapeutic agent, and the administration of such transduced cells to a patient in order to achieve a therapeutic effect.

Moritz teaches the transfection of cord blood cells with retroviral vectors including the neomycin resistance gene, and with retroviral vectors including the adenosine deaminase gene. The cord blood cells also may be cultured in the presence of c-kit ligand and Interleukin-6. Moritz, however, provides no suggestion to one of ordinary skill in the art that CD34+ cells may be separated from the other cord blood cells prior to the transduction of the cells, or that genetically engineered CD34+ cells may be administered to a human patient in order to provide a therapeutic effect.

Kohn discloses the culturing of CD34+ cells obtained from bone marrow in the presence of Interleukin-1, Interleukin-3, Interleukin-6, and human mast cell growth factor, or MGF. The culturing of CD34+ cells in the presence of these growth factors provides for improved retroviral transduction of such cells. Kohn, however, is directed solely to obtaining CD34+ cells from bone

marrow, and does not disclose or suggest to one of ordinary skill in the art that one may genetically engineer CD34+ cells obtained from cord blood, and administer such genetically engineered CD34+ cells to a human patient in order to achieve a therapeutic effect.

Applicants and only Applicants have discovered that because the number of circulating hematopoietic progenitor cells drops to levels seen in older children and adults within two days of birth, collection of cord blood cells at birth enables one to obtain increased quantities of cells such as CD34+ cells, which are useful in gene therapy, in a manner which is safe and efficient. The cited prior art is directed either to the transduction of CD34+ cells which are not obtained from cord blood, or to the transduction of cord blood cells in general, without any suggestion that CD34+ cells may be separated from the other cord blood cells prior to the transduction of the cells. In addition, the cited prior art provides no reasonable expectation that autologous CD34+ cells obtained from cord blood and which are genetically engineered with at least one nucleic acid sequence encoding a therapeutic agent, may be administered to a patient to obtain expression of an effective amount of the therapeutic agent *in vivo*. At best, the cited prior art would render it obvious to try to obtain CD34+ cells from cord blood, genetically engineer such CD34+ cells, and administer such genetically engineered CD34+ cells to a patient to provide a therapeutic effect. The case law has held that such a standard for obviousness clearly is improper. (See American Hospital Supply Corp. v. Travenol Laboratories, Inc., 223 U.S.P.Q. 577 (C.A.F.C. 1984), at 583; Uniroyal, Inc. v. Rudkin-Wiley Corp., 5 U.S.P.Q.2d 1434 (C.A.F.C. 1988), at 1440.) The Examiner also has relied upon the improper use of hindsight gleaned solely from Applicants' disclosure. (See Interconnect Planning Corp. v. Feil, 227 U.S.P.Q. 543 (C.A.F.C. 1985), at 551; In re Dow Chemical, 5 U.S.P.Q.2d 1529 (C.A.F.C. 1988), at 1532; In re Fine, 5 U.S.P.Q.2d 1596 (C.A.F.C. 1988), at 1600.) Because the Examiner's holdings of obviousness is based upon improper standards, Applicants' claimed invention is not rendered obvious to one of ordinary skill in the art by the cited prior art, and it is therefore respectfully requested that the rejection under 35 U.S.C. 103 be reconsidered and withdrawn.

Claims 1-10 and 21-24 stand rejected under 35 U.S.C. 103 as being unpatentable over Anderson (Science) taken with Mitani and Culver. This rejection is respectfully traversed.

The Anderson (Science) reference has been discussed hereinabove. Culver, like Anderson, discloses the transfection of T-cells with a retroviral vector including the adenosine deaminase

gene. The T-cells then were given to children suffering from severe combined immune deficiency. After receiving the transduced T-cells, the children demonstrated a substantial increase in the number of circulating T-cells and an increase in adenosine deaminase activity. Culver, however, provides no suggestion to one of ordinary skill in the art that one may genetically engineer CD34+ cells obtained from cord blood, followed by the administration of such genetically engineered cells to a human patient in order to achieve a therapeutic effect.

Mitani discloses the transduction of a stem cell enriched CD34+ fraction of bone marrow cells *in vitro* with a retroviral vector including the human adenosine deaminase gene. Mitani, however, is directed solely to the *in vitro* transduction of CD34+ cells obtained from bone marrow, and does not even remotely suggest to one of ordinary skill in the art that one may obtain CD34+ cells from cord blood, genetically engineer such cells such that the CD34+ cells include at least one nucleic acid sequence encoding a therapeutic agent, and administering the CD34+ cells to a human patient in order to achieve a desired therapeutic effect. Therefore, Mitani does not render Applicants' invention as claimed obvious to one of ordinary skill in the art.

The combination of Anderson, Culver; and Mitani, at best, teach one skilled in the art to obtain a fraction containing CD34+ cells from bone marrow, and to transduce such CD34+ cells with at least one nucleic acid sequence encoding a therapeutic agent, followed by the administration of such transduced cells to a human patient. Such combination does not even remotely suggest that one may obtain an increased amount of CD34+ cells by obtaining a sample of cord blood from an infant, and separating the CD34+ cells from the cord blood. Thus, one may obtain an increased number of CD34+ cells which may be transduced with at least one nucleic acid sequence encoding a therapeutic agent. The cited prior art provides no basis for obtaining CD34+ cells from cord blood, and genetically engineering such cells with at least one nucleic acid sequence encoding a therapeutic agent, and administering such cells to a human patient. Again, the Examiner has relied solely upon hindsight in order to formulate the holding of obviousness. It is therefore respectfully requested that the rejection under 35 U.S.C. 103 be reconsidered and withdrawn.

Applicants also traverse the rejection of the claims under the judicially created doctrine of obviousness-type double patenting over Claims 1, 2, 8, 9, 10, 13, and 14 of U.S. Patent No. 5,399,346. The '346 patent discloses providing a human with a therapeutic protein by introducing

human cells into a human. The human cells were treated *in vitro* to insert therein a DNA segment encoding a therapeutic protein. The human cells express a therapeutically effective amount of the protein in the human. The '346 patent provides various examples of human cells which may be genetically engineered; however, the '346 patent does not disclose or suggest that autologous CD34+ cells obtained from cord blood may be transfected with a DNA sequence encoding a therapeutic agent, and be returned to the patient, whereby the autologous CD34+ cells express an effective amount of the therapeutic agent *in vivo*. Therefore, the '346 patent provides no basis for an obviousness-type double patenting rejection, and it is therefore respectfully requested that such obviousness type double patenting rejection be withdrawn.

The claims stand rejected under 35 U.S.C. 112, first paragraph, in that the specification fails to provide an enabling disclosure. This rejection is respectfully traversed.

The Examiner has taken the position that one of ordinary skill in the art would not be able to predict that the results shown in Applicants' working examples as being sustained far into the future with any expectation of success.

The Examiner, by his own admission, acknowledges that Applicants have demonstrated the obtaining of CD34+ cells from the cord blood of newborn infants having adenosine deaminase deficiency, and that such CD34+ cells are infected with a retroviral vector including the human adenosine deaminase gene. The infected CD34+ cells then are administered to the infants. The Examiner admits that there are immediate therapeutic benefits, such as the development of normal numbers of T-lymphocytes, normal PHA responses, and normalized levels of deoxyadenosine metabolites. In addition, PCR analyses of peripheral blood samples show that each child has circulating leukocytes which contain the vector-transferred ADA gene. Such analyses demonstrated that there was effective gene transfer into at least some long-lived hematopoietic progenitor cells which are continuing to contribute to peripheral blood cell pools out to at least eight months. Thus, Applicants have demonstrated therapeutic benefits and persistence of the ADA gene in blood cells for at least eight months following treatment, and have proven the principle that one may administer autologous genetically engineered CD34+ cells to a patient and achieve *in vivo* expression of a therapeutic agent. The Examiner has provided no evidence that the administration of CD34+ cells obtained from cord blood are genetically engineered with at least one nucleic acid sequence encoding a therapeutic agent would not result in sustained

therapeutic effects for a term longer than that demonstrated by Applicants. Thus, the Examiner has not shown that one skilled in the art would doubt the asserted utility of Applicants' claimed method. Therefore, Applicants' disclosure corresponds in scope to the claimed subject matter. (See Ex Parte Rubin, 5 U.S.P.Q.2d 1461 (Bd. Pat. App. Int. 1987), at 1462.) Instead of providing substantive evidence which indicates that Applicants' claimed method of treating a human patient would not be successful or effective, the Examiner instead provides mere allegations that one skilled in the art cannot predict the long-term success of Applicants' claimed method, or that one skilled in the art cannot predict the efficacy of a treatment wherein transduced cells are reinfused into a human. Such allegations, in the absence of substantive evidence, are insufficient for maintaining a rejection under 35 U.S.C. 112.

The Examiner also stated that Applicants disclose percentages of colonies of CD34+ cells that had the neo^R gene inserted, such cells being transduced with a retroviral vector including an adenosine deaminase gene and a neomycin resistance gene; however, the Examiner states that Applicants do not teach high transfection of hematopoietic stem cell precursors. In response, Applicants assert that because neomycin resistance selection was employed, high transfection of the stem cell precursors was not required. Applicants, in fact, have demonstrated the genetic engineering of CD34+ cells obtained from cord blood by transducing with cells with a retroviral vector including an adenosine deaminase gene and the administration of such genetically engineered cells to provide a therapeutic effect. Thus, Applicants have enabled the practice of the claimed invention.

The Examiner also states that Applicants' disclosure of culturing transduced CD34+ cells in the presence of Interleukin-3, Interleukin-6, and a c-kit ligand presents further uncertainty because the use of growth factors is still experimental and more detailed understanding of growth factors is needed in the art. The issue is not, as the Examiner asserts, whether the CD34+ cells will expand in culture, but rather whether such cells may be transduced with at least one nucleic acid sequence encoding a therapeutic agent. Applicants have found that if CD34+ cells are cultured in the presence of Interleukin-3, Interleukin-6, and a c-kit ligand, one obtains improved transduction of the CD34+ cells. Again, the Examiner has not provided any evidence which would indicate that such an assertion by Applicants is unbelievable. In addition, the quotation cited by the Examiner from the Kohn reference regarding the role of growth factors in applying

gene transfer into hematopoietic stem cells does not mean that one cannot transduce CD34+ cells with desired genes, but rather that growth factors may be able to improve gene transfer into hematopoietic stem cells. Thus, the Examiner has not met his burden in showing that CD34+ cells could not be cultured in the presence of Interleukin-3, Interleukin-6, and a c-kit ligand and be transfected with at least one nucleic acid sequence encoding a therapeutic agent. Therefore, this embodiment of the claimed invention complies with the requirements of 35 U.S.C. 112.

For the above reasons and others, the specification provides an enabling disclosure, and it is therefore respectfully requested that the rejection under 35 U.S.C. 112, first paragraph, be reconsidered and withdrawn.

The claims stand rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter Applicants regard as the invention. This rejection is respectfully traversed.

The Examiner stated that the claims were vague and seemingly inconsistent in that no relationship exists between the cells that Applicants intend on administering to the patient and the cord blood from where the CD34+ cells are obtained. The claims have been amended in order to define the CD34+ cells as autologous CD34+ cells.

Regarding the Examiner's comment that the claims are vague in that no mention of whether the therapeutic strategy is to be *in vivo* or *in vitro*, the claims define a method of administering genetically engineered CD34+ cells to a patient, whereby the patient is provided with a therapeutic agent. Thus, one skilled in the art would understand readily that the therapeutic agent is being provided to the patient by the CD34+ cells *in vivo*.

Claim 1 has been rewritten to indicate more clearly that the term "administering" means that the CD34+ cells are administered to the patient. With respect to the term "genetically engineered," one skilled in the art would understand readily that such term means any means by which a desired nucleic acid sequence may be introduced into a CD34+ cells, and examples of such means are provided at Pages 3 and 4 of the specification.

In addition, Applicants assert that the term "at least one" also is understood by one skilled in the art, and one skilled in the art could determine whether the administration of CD34+ cells to a patient would be infringing Applicants' Claims 1-5 and 15-26.

Regarding the Examiner's holding that Claim 1 claims a method without disclosing any therapeutic effect, applicants maintain that the therapeutic effect is achieved by providing the patient with the therapeutic agent expressed by the genetically engineered CD34+ cells, and that the specific therapeutic effect is dependent upon the disease or disorder being treated.

Claims 6 and 11 have been amended to define the administration of CD34+ cells including a nucleic acid sequence encoding adenosine deaminase in an amount effective to treat severe combined immune deficiency.

With respect to the Examiner's comments regarding Claim 16, Applicants note that such claim is directed to a method of genetically engineering CD34+ cells. Thus, the genetically engineered CD34+ cells are end product of the process. Such is understandable readily by those skilled in the art, and therefore, Claim 16 complies with the requirements of 35 U.S.C. 112, second paragraph.

For the above reasons and others, the claims particularly point out and distinctly claim the subject matter Applicants regard as the invention, and it is therefore respectfully requested that the rejection under 35 U.S.C. 112, second paragraph, be reconsidered and withdrawn.

For the above reasons and others, this application is in condition for allowance, and it is respectfully requested that the rejection be reconsidered and withdrawn and a favorable action is hereby solicited.

Respectfully submitted,

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Name of applicant, Assignee, or Registered Representative

Signature

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Date of Signature